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NEPHROLOGY FORUM

Immunologic and genetic aspects of systemic lupus erythematosus

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Case presentation

A 38-year-old woman was admitted to the New England Medical Center Hospital (NEMCH) 4 years ago with a 10-year history of intermittent fatigue, arthralgias, oral ulcers, pleurisy, and a photosensitive facial rash. Blood pressure was 140/80 mm Hg, and physical examination revealed a malar rash, peripheral adenopathy, and splenomegaly. Serologic studies revealed an anti-DNA titer of 24. mg/ml (normal < 2.5 mg/ml), a total serum hemolytic complement (CH_{50}) of 58 units (normal > 50 units), and a C3 of 0.32 mg/ml (normal .87 to 2.2). Urinalysis showed 3 + protein; 1 + blood; and 10 to 15 white blood cells and 2 to 4 red blood cells per high-power field. The 24-hour urine protein excretion was 1.3 g. Serum creatinine was 0.8 mg/dl.

The patient was treated initially with 40 mg of prednisone daily, but the serum complement levels remained depressed. Her blood pressure rose to 180/110 mm Hg, and treatment with hydrochlorothiazide was initiated. She tolerated prednisone poorly, developing mood changes and Cushingoid features. The dose of prednisone was gradually reduced to 10 mg per day over a 6-month period. She continued to have intermittent pleuritic chest pain and arthralgias. Three and one-half years ago treatment with azathioprine was initiated at a dose of 100 mg/day.

Approximately 3 years ago, the serum creatinine was 1.7 mg/dl. Urinalysis showed 2+ protein with 5 to 6 white blood cells and 10 to 20 red blood cells per high-power field. Prednisone was increased to 60 mg per day. Diastolic blood pressures remained in the range of 110 to 120 mm Hg; propranolol, hydralazine, and furosemide were added to her therapeutic regimen. The dose of azathioprine was increased transiently to 150 mg per day but had to be lowered to 50 mg per day because of leukopenia. During the ensuing 6 months, the serum creatinine rose progressively to 2.7 mg/dl and the 24-hour urinary protein excretion increased to 6 to 7 g. By the end of that 6-month period, azathioprine was discontinued and the patient was treated with 60 mg of cyclophosphamide.

She was readmitted to the NEMCH 2.5 years ago with a blood pressure of 230/130 mm Hg and anasarca. The serum creatinine was 1.9 mg/dl, the 24-hour urine protein excretion was 10.4 g, and the serum albumin was 2.4 g/dl. Blood pressure was controlled with the addition of alpha-methyldopa and intravenous furosemide to the therapeutic regimen. Prednisone was increased to 100 mg per day. Cyclophosphamide was discontinued because the anti-DNA titer and CH_{50} had returned to the normal range. A percutaneous renal biopsy was performed.

During the subsequent 3 months, the patient developed a severe proximal myopathy, marked Cushingoid features, and osteoporosis complicated by a thoracic vertebral compression fracture. The dose of prednisone was gradually tapered again. For the next 2 years, her blood pressure was well controlled despite an intolerance to many antihypertensive medications and the patient's noncompliance in taking them. Renal function remained stable; serum creatinine ranged from 2.0 to 2.5 mg/dl. Anti-DNA titers remained mildly elevated but serum complement levels generally remained in the normal range. One year ago, prednisone was tapered to 20 mg every other day; however, treatment with azathioprine was reinstituted because of persistent arthralgias.

She was readmitted to the NEMCH 6 months ago because of deteriorating renal function and poorly controlled hypertension. Her blood pressure, which was 240/180 mm Hg on admission, was controlled by treatment with minoxidil. The serum creatinine was 3.5 mg/dl. Urinalysis showed microscopic hematuria and many red blood cell casts. Prednisone was again increased to 80 mg per day. Azathioprine could not be increased beyond 50 mg per day because of persistent leukopenia. During the next 4 months, her renal function deteriorated further despite 5 one-liter plasma exchanges performed over 2 weeks. Although high-dose prednisone therapy was continued, she required hospitalization on two later occasions because of acute abdominal pain secondary to lupus peritonitis. Myopathy recurred and another vertebral compression fracture developed. Two months ago, the serum creatinine was 8.5 mg/dl, uremic symptoms appeared, and

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maintenance hemodialysis was initiated. Treatment with azathioprine was discontinued and the dose of prednisone was again tapered. She continues to require minoxidil and propranolol for control of hypertension.

Discussion

DR. ROBERT S. SCHWARTZ *(Chief, Hematology-Oncology Division, New England Medical Center Hospital; Director, Cancer Research Center, and Professor of Medicine, Tufts University School of Medicine, Boston, Massachusetts):* A kidney biopsy was performed shortly after this patient's renal function began to deteriorate. What were the pathologic findings?

DR. VIVIAN PINN *(Associate Professor of Pathology, Tufts University School of Medicine, Boston, Mass.):* The renal biopsy specimen contained approximately 30 glomeruli, all of which were involved by a moderate to severe degree of mesangial proliferation. In many instances the entire glomerulus or a segment of it had a lobular appearance (Fig. 1A). Occasional polymorphonuclear leukocytes were evident within glomerular tufts, and about one-third of the glomeruli also had epithelial cell proliferation with localized crescent formation. Many intracapillary thrombi were present. The glomerular capillary basement membrane was irregularly thickened and some areas exhibited a "wire-loop" configuration. Foci of interstitial chronic inflammatory infiltrates also were present.

Immunofluorescent studies demonstrated irregular but prominent fine to coarse granular deposition of immunoglobulins (IgG, IgM, IgA) with a similar pattern of deposition of C3, C4, and C1q, predominantly on the peripheral capillary basement membrane but also in the mesangium and foci of Bowman's capsule (Fig. 1B). Granular deposits of IgG and C3 also were identified on the tubular basement membrane.

Electron microscopy confirmed the findings by light microscopy and showed prominent subendothelial and mesangial electron-dense deposits with scattered intramembranous and rare intramembranous deposits (Fig. 1C). Mesangial cell proliferation with increased mesangial matrix often extended along the peripheral capillary basement membrane and produced a double contour appearance similar to that seen in membranoproliferative glomerulonephritis. Epithelial cells were hypertrophied with effacement of foot processes. In summary, the biopsy demonstrated diffuse proliferative lupus nephritis with a membranoproliferative glomerular pattern.

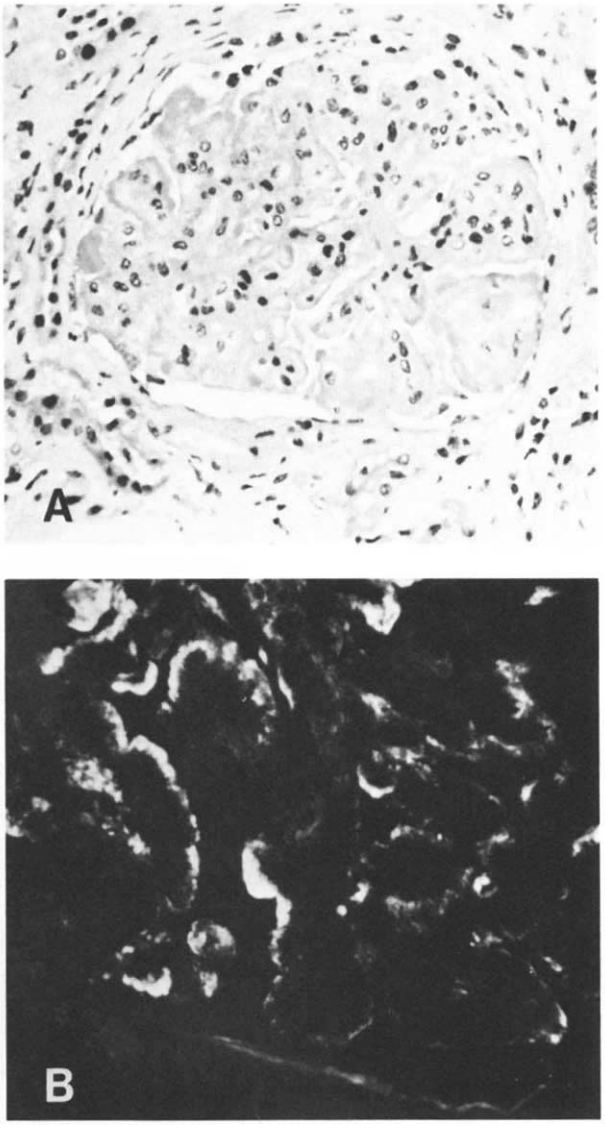

Fig. 1 A *Light microscopy of a glomerulus demonstrating the accentuated lobular architecture, increased mesangial cells and matrix, and thickened peripheral capillary loops* (×250, hematoxylin and eosin). **B** *Photograph of immunofluorescent demonstration of IgG deposition in an irregular granular distribution along peripheral glomerular capillary basement membranes, Bowman's capsule, and the mesangium* (×600).

DR. SCHWARTZ: This woman's medical course and her biopsy illustrate a classic example of extensive immunologic injury in a patient with systemic lupus erythematosus (SLE). The autoimmune disease par excellence, SLE is a disorder with many challenges for both clinicians and research scientists. The clinician must deal with patients whose disease can affect any organ, from brain to bowel; the researcher seeks to unravel the immunologic, genetic, hormonal, and environmental factors that

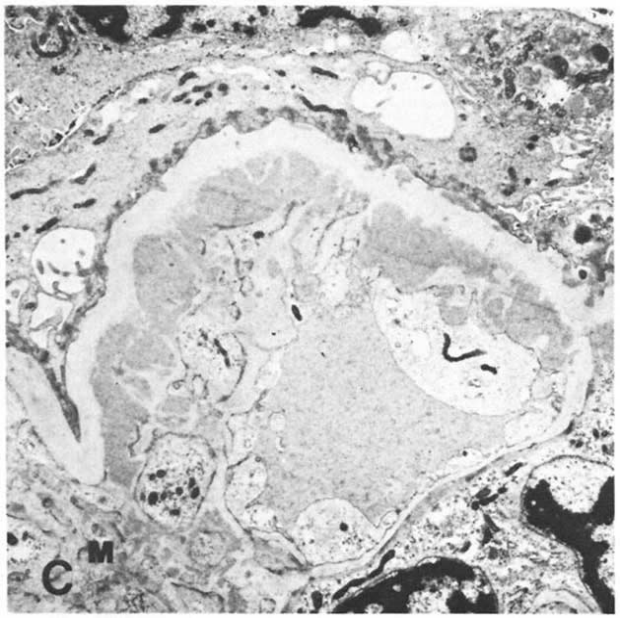

Fig. 1 C *Electron micrograph of a glomerular capillary loop with prominent subendothelial and mesangial electron-dense deposits as well as sparse subepithelial deposits* (M = mesangium). Subendothelial mesangial interposition can be seen at the periphery of the capillary loop (×2800).

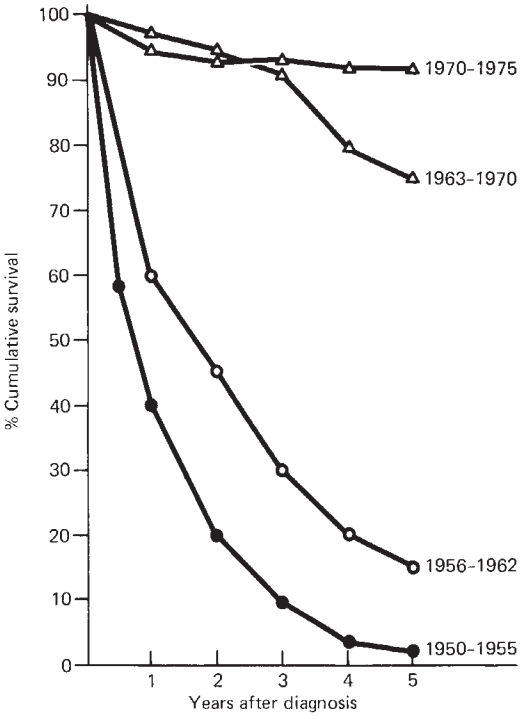

Fig. 2. *Survival in SLE, 1950–1975.* Data obtained from Dubois [1] and Fries and Holman [2]. Improved diagnostic tests have led to the inclusion of earlier and milder cases of SLE, thus adding to the improved survival.

form a matrix of causative events in the disorder. The clinician must be prepared to distinguish organic brain disease from a psychiatric disorder, recognize a variety of dermatologic manifestations, treat hypertension, decide about splenectomy, and deal with the emotional upheavals of chronically ill patients who require treatment with potentially toxic drugs. Researchers must use the concepts and tools of cellular immunology, immunochemistry, immunopathology, genetics, and even virology and endocrinology. Thus, SLE is not a disease for narrow specialists.

Despite the complexity of SLE, considerable advances have been made, especially during the past decade. Data from Dubois [1] and from Fries and Holman [2] provide the most convincing evidence of this progress, that is, the dramatic improvement in prognosis (Fig. 2). In 1950, virtually all patients with SLE died within 5 years of diagnosis; today, only about 10% succumb to the disease within that time. This remarkable change is due to: (*1*) general improvements in medical care since 1950; (*2*) the introduction of specific and sensitive diagnostic tests for SLE, which have made it possible for us to recognize the disease in its early stages; and (*3*) increased sophistication in the application of corticosteroid and immunosuppressive therapy. Nevertheless, important clinical problems remain. Involvement of the central nervous system and the kidney continue to be major causes of disability and death. Diffuse lupus nephritis is still an ominous development, and the outlook for patients with this complication is as poor today as was the general prognosis for patients with SLE 25 years ago [3] (Fig. 3).

Several excellent papers offer detailed reviews of the clinical and serologic features of SLE [4–6]. I will focus, therefore, on the causative matrix, which is the centerpiece of current research in SLE. We will examine the idea that SLE is a genetic abnormality of the immune system and that SLE is a *syndrome* whose manifestations can arise from different types of genetic abnormalities, all of which ultimately lead to a common pathogenetic mechanism, namely the deposition of circulating immune complexes in various tissues.

The best evidence that SLE has a genetic basis is the high concordance of the disease in monozygotic twins. Block et al reviewed the cases of 63 patients, each of whom had an identical twin [7]. The concordance of the disease in this group was 63% (40 of 63), a rate that indicates a strong genetic influence. This figure of almost two-thirds is actually a mini-

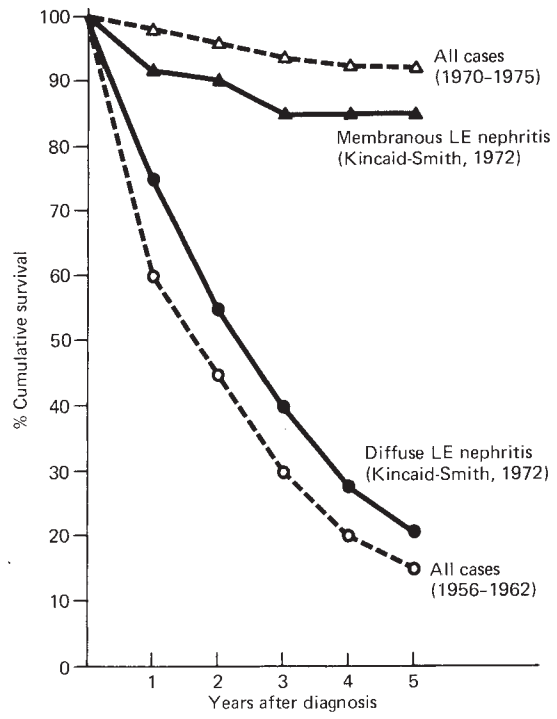

Fig. 3. *Influence of lupus nephritis on the prognosis of SLE.* Lines labeled "Kincaid-Smith" were derived from Ref. 3.

mal estimate; the onset of the disease in the twins was temporally discordant, and additional cases of concordant disease thus are expected. Another interesting aspect of this survey was the confirmation of a relatively high incidence of serologic abnormalities in healthy, first-degree relatives of patients with SLE (Table 1). The difference between spouses and first-degree relatives in this regard is highly significant ($\chi^2 = 59.5$, $P < 0.001$) and points to another kind of genetic influence to which I shall return.

When the disease occurs in twins, clinical manifestations tend to be similar in both [7]. In light of the large number of possible clinical expressions of the disease, this tendency seems remarkable. Another set of genetic factors thus appears to influence the particular way in which patients express the disease. From these observations, we can conclude that at least three kinds of genetic factors influence SLE: those that result in clinically overt disease; those that result in a tendency toward the formation of autoantibodies, but not toward the disease; and those that dictate the actual lesions of the disease.

Genes cannot explain SLE entirely, however. If this were so, the concordance rate in identical twins would be 100%. Also, it is well known that females develop SLE more than males do: a survey of almost 1000 patients with SLE disclosed that 90% were females and that the median age was 28.4 years [1]. Because SLE is a disease of young women at the peak of their reproductive capacities, we can deduce that sex hormones profoundly influence the disease. Experiments from Talal's group support this conclusion [8, 9]. He and his colleagues have shown in an animal model of SLE that administration of estrogens accelerates the development of lupus nephritis, whereas androgens hinder its development. Whether these hormones act on autoantibody-producing cells or whether they influence the handling of immune complexes by the reticuloendothelial system is unknown. However the effects of these hormones are explained, it is clear that they affect genetic mechanisms in an important way. The influence of estrogens on gene expression is, of course, a well-known phenomenon [10, 11].

The clinical manifestations of SLE are sometimes not distinctive, but the serologic abnormalities are its hallmark (Table 2). The patient with SLE produces numerous kinds of autoantibodies: the most characteristic of these are the antibodies against nucleic acids, notably antibodies to native and dena-

Table 1. Evidence for genetic factors in SLE[a]

	ANA[b]	SLE
Monozygotic twins	41/63 (65%)	40/63 (63%)
First-degree relatives	182/1113 (16%)	1/53 (2%)
Spouses	36/762 (5%)	0/762 (0%)

[a] Data from Ref. 7.
[b] ANA refers to a positive antinuclear antibody test.

Table 2. Partial list of autoantibodies found in SLE

Against components of cell nuclei
Native DNA
Denatured DNA
Nucleoprotein
Single-strand RNA
Double-strand RNA
Sm antigen
Oligonucleotides
Against components of cytoplasm
Ribosomes
Lysosomes
Ro antigen
Against cell membranes
Red cells
Platelets
Lymphocytes
Granulocytes
Other
Cardiolipin
Coagulation factors

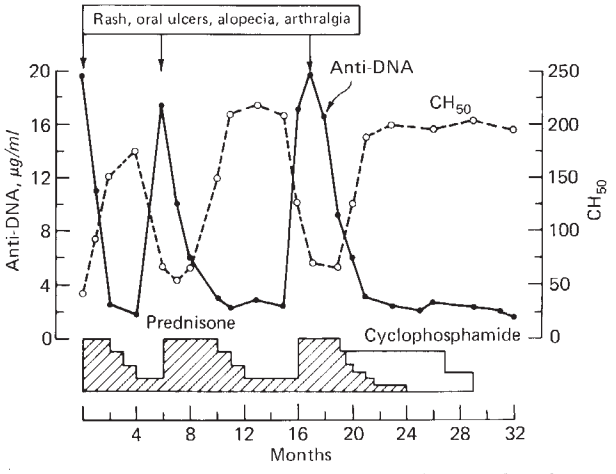

Fig. 4. *Clinical chart of a patient with SLE showing the relationship between activity of the disease and serologic abnormalities.* The inverse relationship between levels of anti-DNA antibodies (solid line) and levels of hemolytic complement activity (dashed line) is noteworthy.

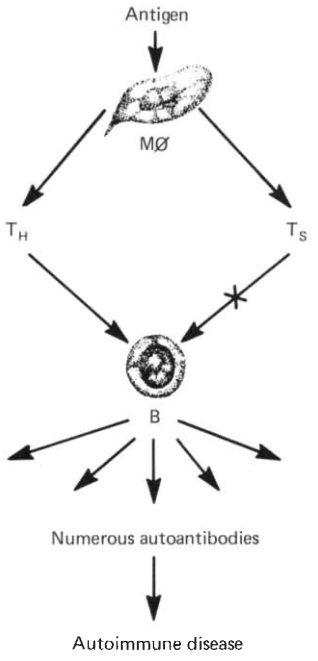

Fig. 5. *Suppressor cell theory of SLE.* T_H refers to helper T cell, T_S refers to suppressor T cell. The lines and arrows represent regulatory loops in schematic form. According to this theory, defective T_S function "unleashes" B cells.

tured DNA. Studies of these antibodies can illustrate four important aspects of lupus. First, their presence is a highly specific diagnostic finding. Antibodies that bind to native DNA or to the nucleoprotein Sm antigen are rarely found except in patients with SLE [12]. Second, some of the serologic abnormalities provide useful markers by which the activity of the disease can be measured objectively, and perhaps even predicted [13]. Antibodies to DNA and levels of complement activity in the blood are particularly useful in this regard (Fig. 4). A third and related aspect is the abundant evidence that these autoantibodies participate in the pathogenesis of the lesions of SLE. For example, anti-DNA autoantibodies are important constituents of the immune complexes that deposit in the glomerular basement membranes [4]. Fourth, and currently the most perplexing aspect of lupus, is the explanation of how these antibodies originate. It is to this question that I would now like to turn.

Contemporary cellular immunology can be summarized in two words: recognition and regulation. "Recognition" refers to those mechanisms whereby lymphocytes recognize not only foreign structures, but each other; "regulation" refers to mechanisms that control the immune response. These definitions are useful because the cytologic simplicity of lymphocytes masks the complexity of their function. Look-alike lymphocytes actually comprise various subsets, each with different functions. Three major divisions exist among these subsets: helper T lymphocytes, suppressor T lymphocytes, and B lymphocytes. These kinds of cells, together with macrophages, constitute the primary elements of the immune system. They are held together in a functional unit by a network of regulatory signals in a manner analogous to the web of signals that integrates the cells of the brain. The lymphocytic signals, whether they extend from cell surfaces or diffuse through extracellular fluids, are responsible for both recognition and regulation.

A predominant theory of autoimmunization proposes that impaired regulation of the immune system leads to the production of autoantibodies [14]. This theory assumes the existence of B lymphocytes that have the potential to produce autoantibodies. This potential normally is not realized because of the restraining influence of immunoregulatory T lymphocytes. According to the most popular version of this theory, a qualitative or quantitative defect of suppressor T cells would fail to regulate potential autoantibody-producing B lymphocytes, thereby "unleashing" them (Fig. 5).

One recent theory does not require defective immunoregulation for autoimmunization. Instead, the primary disturbance is thought to involve clones of B lymphocytes that produce autoantibodies despite normal regulatory signals from T lymphocytes [15]. These B lymphocytes are thus "outlaws" (Fig. 6).

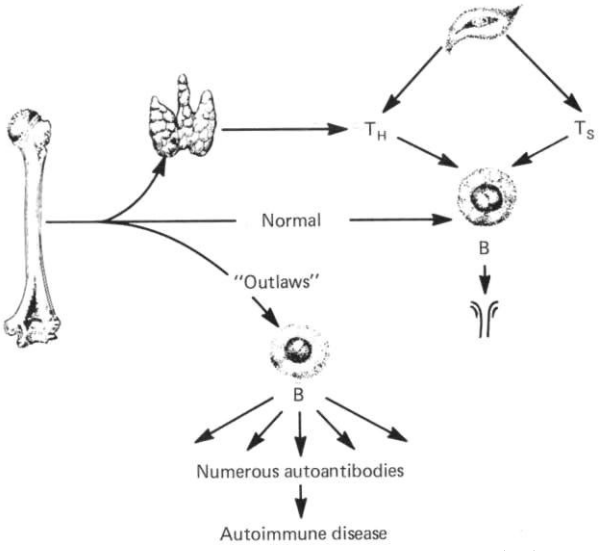

Fig. 6. *Outlaw B cell theory of SLE.* According to this theory, a subset of intrinsically abnormal B cells fails to respond to immunoregulatory signals and produces autoantibodies.

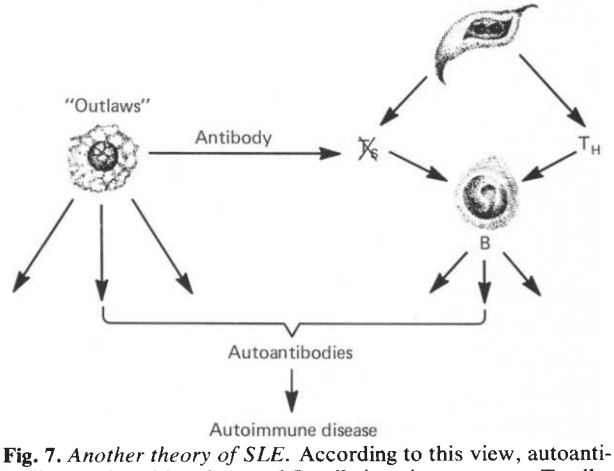

Fig. 7. *Another theory of SLE.* According to this view, autoantibodies produced by abnormal B cells impair suppressor T cells, thereby accelerating the tendency toward autoimmunization.

Recently, Gershwin and his colleagues at the University of California at Davis showed that severe lupus nephritis developed in the offspring of New Zealand black and New Zealand white mice [(NZB × NZW)F_1] that were homozygous for the gene *nu* (personal communication). Homozygosity for this gene causes congenital absence of the thymus and, as a consequence, the absence of any mature T cells. Thus, autoimmunization with the production of a fatal form of lupus nephritis can occur in animals whose functioning lymphocytes consist almost entirely of B cells.

A reconciliation between these contrasting theories of autoimmunization is shown in Figure 7 and is based on the finding of autoantibodies to T lymphocytes, particularly autoantibodies specific for suppressor T lymphocytes in SLE [16]. These autoantibodies, which could be produced by intrinsically abnormal ("outlaw") B lymphocytes, might impair immunoregulatory circuits and thereby recruit into the pathogenetic mechanism additional autoantibody-producing B lymphocytes. A vicious cycle arising out of such an immunologic whirlwind can be envisioned.

Solid data from experiments in animal models of SLE support each of these theories. The importance of such animal models to our current understanding of the disease cannot be overemphasized. The most familiar of them is the NZB mouse, a highly inbred strain in which a lupus syndrome regularly develops. Recent studies of NZB mice have shown that their B lymphocytes spontaneously secrete large amounts of immunoglobulins [17]. This so-called spontaneous polyclonal activation of B cells can be detected even in fetal NZB mice and occurs independently of any known T-lymphocyte influence [18]. Not only is immunoglobulin synthesis increased markedly, but the number of immunoglobulin-containing cells is also increased. Furthermore, the amount of immunoglobulin secreted per cell is higher than that found in B lymphocytes of normal mice. A separate genetic mechanism dictates each of these abnormal properties of NZB immunoglobulin-producing cells [18]. For instance, the number of immunoglobulin-containing cells is a dominant autosomal trait, whereas the increased secretion rate per cell is controlled by autosomal recessive genes. The abnormal cells represent a distinct and minor population, which has been traced to a particular subtype of B lymphocytes [19]. The remaining B lymphocytes in NZB mice, by contrast, seem normal.

Andrews et al have investigated the clinical and serologic manifestations of SLE in several other experimental models [20] (Table 3). Each of these inbred lines of mice has distinct immunologic, genetic, and hormonal features. Yet each develops one or more characteristic lesions of SLE. These various models add to the evidence that SLE is a syndrome that can result from one of several different pathways. Each inbred strain represents a genetic "window" through which one of the pathways to lupus can be perceived. It thus would be surprising if a single mechanism could explain SLE in a genetically heterogeneous human population. There

Table 3. Clinical and serologic findings of SLE in 6 highly inbred strains of mice[a]

Manifestation	Inbred strain					
	NZB	B/W	MRL/1	BXSB	Me/me	Swan
Hemolytic anemia	+++	0	0	++	0	0
Nephritis	±	+++	++++	+++	+	++
Vasculitis	+	±	++	0	0	0
Dermatitis	0	0	++	0	+	++
Arthritis	0	0	++	0	0	0
Anti-DNA antibodies	++	+++	++++	+	−[b]	−
Anti-Sm antibodies	0	0	++	0	−	−
Anti-lymphocyte antibodies	+++	+++	++	+	++	−
Rhematoid factor	0	±	++	0	−	−

[a] Each genetically uniform strain presents a distinct lupus syndrome. Approximate severity (clinical or pathologic) or levels of autoantibodies are estimated on a scale from 0 to ++++. Data from Ref. 20.

[b] Indicates unknown.

is, therefore, no place for dogmatism in formulating notions about the etiology of SLE.

None of the current ideas, however, comes to grips with the serologic abnormalities of SLE. None of them explains exactly how the patient forms antibodies to DNA. Indeed, all of the current theories assiduously avoid any mention of this specific feature of the disease. This neglect is not without cause: little is known about the immunochemical specificities of the autoantibodies in SLE. Our ignorance of this aspect of SLE stems from the fact that lupus serum is a complex mixture of antibodies. No method is available for separating this mixture into its component parts so that a single molecular species of autoantibody can be analyzed. Recently, however, a powerful technique with exciting possibilities for studies of individual antibodies was devised by Köhler and Milstein [21]. This method utilizes somatic cell hybridization to immortalize single antibody-forming cells by fusing them with myeloma cells. Such hybridomas, as they are called, can be cloned and, after either in vitro or in vivo culture, they produce monoclonal antibodies that are encoded by genes derived from the antibody-producing B lymphocyte partner of the fusion product. The Köhler and Milstein method has revolutionized immunology because of its analytic power and its enormous specificity.

My colleagues and I have begun to apply "hybridoma technology" to a study of spontaneously produced autoantibodies to DNA. For this purpose, we fused spleen cells from unimmunized MRL/1 mice with mouse plasmacytoma cells. The MRL/1 mice develop a severe form of lupus and produce high levels of autoantibodies to DNA (see Table 3). The fusion procedure led to the isolation of cloned hybridomas that secrete single molecular species of autoantibodies to DNA [22]. These hybridoma autoantibodies also have been used to prepare antiidiotype antibodies in rabbits. "Idiotype" refers to a structure in the hypervariable region of an immunoglobulin molecule that constitutes a portion of its antigen-binding domain. Such structures would be specific for a given antibody molecule, and thus they represent "markers" of exceptional specificity. The antiidiotypic antisera provide the means by which these markers can be detected and quantitated.

These new reagents already have yielded two important findings [23]. The first is that monoclonal autoantibodies to DNA can manifest multiple serologic specificities. For instance, many of the cloned "anti-DNA" antibodies also bind to several different polynucleotides, including poly(I), poly(G), and poly(A). This surprising behavior, unexpected from a single molecular species of antibody, implies the existence of shared antigenic structures among a wide variety of nucleic acid antigens; such structures might reside within the sugar-phosphate backbones of DNA and other polynucleotides. This finding also implies that the number of autoantibodies produced in SLE is less than the number suggested by the many serologic manifestations of serum from patients with lupus, because a single species of autoantibody can have diverse serologic properties.

The second important finding revealed by the antiidiotype reagents is the presence of similar idiotypes among different monoclonal autoantibodies from different individuals. The genes that encode these crossreacting structures therefore might share certain nucleotide sequences. One implication of this finding is that "families" of related autoantibody-encoding genes might exist in the germ line of MRL/1 mice. If this interpretation is borne out, the fundamental defect in SLE will boil down to a defect of gene regulation.

Some researchers have used a Mendelian approach to study the genetics of SLE. Such studies examine how the phenotypes of the disease segregate among crosses between affected and normal individuals. The (NZB × NZW)F_1 hybrid is a striking example of the power of this orthodox approach. Although NZB mice regularly develop autoimmune hemolytic anemia as part of their lupus complex, nephritis is rare (Table 3). In striking contrast, severe lupus nephritis develops uniformly in the prog-

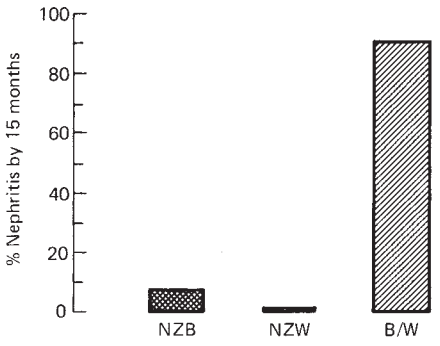

Fig. 8. *Development of lupus nephritis in (NZB × NZW) F_1 hybrid mice (B/W).* Neither the NZB nor the NZW parent develops this disorder. NZB mice have a lupus diathesis, whereas NZW mice are normal and do not produce autoantibodies or form lesions of lupus or any related disorder.

eny of crosses between NZB and NZW mice (a strain in which lupus never occurs) (Fig. 8) [24]. A similar event occurs in crosses between NZB and SWR mice, another normal inbred strain in which neither autoantibodies nor lupus lesions ever develop [25].

These findings signify that genes possessed by normal individuals must influence the clinical expression of the lupus "diathesis." By means of traditional Mendelian techniques, Knight and Adams demonstrated in the (NZB × NZW)F_1 model that three genes (or gene complexes) participate in the development of lupus nephritis, and that two of these derive from the normal NZW parent [26]. These facts have led to the formulation of a new theory about the genetics of SLE [27], which proposes the existence of two families or classes of genes that might be unlinked in the Mendelian sense. Each family, however, encompasses a set of functionally related genes (Table 4). The first of these classes of genes controls the central immune functions of recognition and regulation. It would encompass, for instance, genes that control suppressor and helper T-cell function and HLA genes. The second class of genes specifies the peripheral arm of the immune system, such as immunoglobulin class or avidity, the ability to form soluble immune complexes, and the complex set of mechanisms that comprise the inflammatory response.

Table 4. Dual gene hypothesis of autoimmune disease

Class I genes (central immune function)
Self recognition
Immune response genes
Immunoregulation (helper and suppressor cells)
Class II genes (peripheral manifestations of immunity)
Immune complexes
Complement
Inflammatory response
Class I genes alone: Autoantibodies, no disease
Class II genes alone: Clinically silent (healthy)
Class I + Class II genes: autoimmune disease (e.g., SLE)

Individuals with permissive alleles of only class I genes could form autoantibodies, but they would develop no lesions. These individuals would correspond to the healthy autoantibody-producing, first-degree relatives of patients with SLE, alluded to previously. Individuals with permissive alleles of only class II genes would be healthy. They would be, for instance, the counterparts of NZW or SWR mice. The patient with SLE inherits permissive alleles of both classes of genes. The clinical manifestations of the disease, according to this theory, would result principally from the actions of particular class II genes.

Dr. Kenneth Miller and I tested these ideas experimentally in human SLE [28]. We sought evidence for the existence of permissive alleles of class I genes by a family study in which we examined suppressor T-cell function in first-degree relatives of patients with SLE. The assay system was based on the ability of the plant lectin concanavalin A to stimulate the functional development of suppressor T lymphocytes from a pool of precursor cells. We assayed these lectin-stimulated suppressor cells in turn for their ability to suppress the synthesis of IgG by B lymphocytes. The results demonstrated an impairment of suppressor T-cell function in 13 of 51 clinically healthy, first-degree relatives of patients with SLE. Also, 12 of these 13 relatives were women. This finding supports the view that defective immunoregulation is a genetic abnormality in SLE and also supports the argument that defective immunoregulation cannot *by itself* explain the disease, as the first-degree relatives with the abnormality had no signs of SLE. We presume that other factors, such as permissive alleles of class II genes, must participate in the full expression of this condition.

Our present view of SLE is summarized in Figure 9, which shows the interactions between genetic factors and the environment in the causation of this disorder. One may add to this scheme the role of sex hormones, but the locus of their action is unknown. We can conclude safely that the cause and pathogenesis of SLE is better understood today than ever before. The application of new methods, such as hybridoma technology, to the analysis of this disease holds great promise not only because it ultimately will reveal the molecular biology of

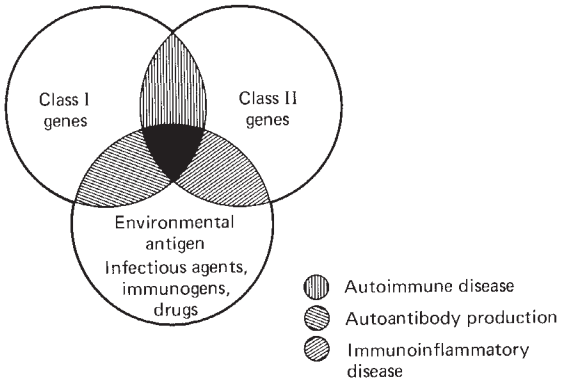

Fig. 9. *Interactions among genes and environmental factors in SLE.*

lupus, but also because the structural requirements for pathogenicity among autoantibodies will be deciphered. This new knowledge, in turn, cannot fail to influence the day-to-day clinical management of the patient with systemic lupus erythematosus.

Questions and answers

DR. JORDAN J. COHEN: Do the renal lesions that occur in NZB/NZW mice conform to the classification scheme of renal disease in patients with SLE?

DR. SCHWARTZ: The usual lesion is diffuse lupus nephritis.

DR. COHEN: Do all the mice develop diffuse lesions or is there a spectrum including membraneous, focal, and other types of nephritis?

DR. SCHWARTZ: I will ask Dr. Janine André-Schwartz whether lesions other than diffuse ones occur in B/W mice.

DR. JANINE ANDRÉ-SCHWARTZ: *(Hematology-Oncology Division, NEMCH):* Yes. Limited focal lesions have been described. Also, limited alterations affecting either the glomerular basement membrane and/or epithelial and endothelial cells can be seen.

DR. COHEN: Is it reasonable to conclude from the wide spectrum of renal lesions that occur in this simple genetic model of SLE that the heterogeneous pattern of renal involvement observed clinically is not necessarily an expression of genetic heterogeneity? A related question has to do with the identical twins who develop SLE with renal involvement. Does a given set of twins necessarily develop the same kind of renal disease?

DR. SCHWARTZ: I don't know the answer to the second question. It is a very interesting point and should be looked into. As to the first question, I would have to say that although the animal models might seem genetically simple, they are in fact complex. Considerable evidence indicates that numerous genes are involved in both the immunologic abnormalities and the pathogenesis of the lesions. For example, there are probably at least six genes that determine the spontaneous hypersecretion of IgM by B cells of NZB mice (unpublished observations). We are attempting to analyze the genetics of the antibodies in the renal lesions with antiiodiotypic antibodies. By this means we will try to estimate how many families of antibodies are involved and the nature of their binding specificities.

DR. COHEN: Are you suggesting that the morphologic classification scheme that we currently use in clinical diagnosis might be homologous with a classification structure based on the nature of the idiotypic antibodies involved in the pathogenesis of the renal damage?

DR. SCHWARTZ: It is too early to say that. I think the present pathologic scheme is an excellent one for clinical use because of its bearing on prognosis.

DR. COHEN: What is your hypothesis about the heterogeneous nature of the renal lesions that occur in patients who have lupus? Is it just a question of how many immune complexes are deposited, or do other factors explain why some patients develop florid renal disease, others only moderate damage, and others none at all?

DR. SCHWARTZ: Certainly the amount of immune complexes deposited in the glomerulus must be a factor. The immunoglobulin class also must be a factor because some classes and even subtypes of IgG fix complement, whereas other classes or subtypes do not fix complement efficiently. Another factor is the size of the immune complex. In experimental models of serum sickness, large immune complexes (those with a sedimentation coefficient of 19) do not deposit in the kidney because they are swept out of the circulation by the spleen and the liver. By contrast, smaller complexes remain in the circulation longer and are deposited more efficiently on the glomerular basement membrane. The avidity of the antibody for antigen also must be a factor. An equilibrium relationship exists between antigen and the corresponding antibody; therefore those antibodies that are highly avid for antigen must be more prone to form stable immune complexes than are less avid antibodies. The hypothesis I presented is that each of these elements that contribute to the lesions of lupus is under genetic control.

DR. JEROME P. KASSIRER: Is it likely that we might be able to identify, either through genetic or

immunologic testing, which patients are more susceptible to serious complications and thus be in a position to treat them earlier?

DR. SCHWARTZ: That is a possibility, but it is difficult to answer your question in a specific way. Much is known about the mechanism of immune complex deposition in the kidney, but what remains unknown is the connection between those factors and genes.

DR. KASSIRER: In Dr. Miller's familial study, were there any hints about genetic transmission of the particular kind of lupus that affected the patient?

DR. KENNETH MILLER *(Hematology-Oncology Division, NEMCH)*: No evidence for this was found.

DR. ANDREW LEVEY *(Renal Fellow, NEMCH)*: Dr. Schwartz, given the genetic insights that you have offered us, what do you mean when you say some patients with SLE have been cured?

DR. SCHWARTZ: It is analagous to saying a patient who has a genetically determined cancer has been cured. Let me stress again that we have to think of lupus not only in genetic terms but also in terms of hormones and the environment. These latter elements might be of considerable importance in provoking the disease in a genetically susceptible person; in this regard, SLE is similar to other genetic diseases in which environment can contribute to their manifestation. Deficiency of glucose-6-phosphate dehydrogenase is a good example.

DR. COHEN: I assume that you would respond similarly to a question about the episodic nature of the clinical manifestations of SLE. Is it true that the clinical manifestations can appear and resolve because of certain nongenetic factors?

DR. SCHWARTZ: Yes.

DR. AARON SPITAL *(Renal Unit, Rhode Island Hospital, Providence, Rhode Island)*: Much controversy exists about the use of immunosuppressive drugs for the treatment of lupus nephritis in view of their obvious side effects. Indeed, today's patient is a good illustration of these sequelae. I think many people would have stopped immunosuppressive treatment a long time before it was discontinued in this patient. Do we know from animal models of this disease whether these drugs alter the natural history of lupus nephritis?

DR. SCHWARTZ: Yes. The (NZB × NZW)F_1 hybrid mouse, which uniformly develops lupus nephritis, has been tested extensively with cyclophosphamide and several other compounds, most of which are therapeutically effective in this model [29]. However, cyclophosphamide and azathioprine have been shown to produce lymphomas in this mouse model [30]. Information obtained from studies of chemotherapeutic agents in the (NZB × NZW)F_1 mouse has not been particularly useful from the clinical point of view because, ironically, such studies have been uniformally successful. This point relates to Dr. Cohen's earlier question about the uniformity of the renal lesion in such inbred animals. Perhaps this fact explains why such treatment is highly successful in inbred mice, but why doubts linger even today about its efficacy in genetically heterogeneous humans.

DR. COHEN: Do you have any thoughts about the role of plasmaphoresis in the treatment of lupus nephritis?

DR. SCHWARTZ: Unless a prospective, controlled, and rigorously designed clinical trial is performed to evaluate the efficacy of plasmaphoresis, we will be faced with yet another unproven mode of treatment for this disease and, in this case, an exceedingly expensive one.

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